

**EFFECTS OF ENERGY DRINK CONSUMPTION ON PHYSICAL
PERFORMANCE AND PERCEPTUAL RESPONSES AMONG
PHYSICALLY INACTIVE UNIVERSITY STUDENTS**

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PERFORMANCE AND PERCEPTUAL RESPONSES AMONG
PHYSICALLY INACTIVE UNIVERSITY STUDENTS**

by

CHAN MING SZE

**Dissertation submitted in partial fulfilment of the requirements for the degree of
Bachelor of Health Science (Honours) (Exercise and Sports Science)**

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CERTIFICATE

This is to certify that the dissertation entitled “ Effects of energy drink consumption on physical performance and perceptual responses among physically inactive university students” is the bona fide record of research work done by Chan Ming Sze during the period from September 2020 to July 2021 under my supervision. I have read this dissertation and that in my opinion it confirms to an acceptable standards of scholarly presentation and is fully adequate, in scope and quality, as a dissertation to be submitted in partial fulfilment for the degree of Bachelor of Health Sciences (Honours) (Exercise and Sports Science).

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DECLARATION

I hereby declare that this dissertation is the result of my own investigations, except where otherwise stated and duly acknowledged. I also declare that it has not been previously or concurrently submitted as a whole for any other degrees at Universiti Sains Malaysia or other institutes. I grant Universiti Sains Malaysia the right to use the dissertation for teaching, research, academic and promotional purposes.



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LIST OF ABBREVIATIONS AND ACRONYMS

%	Percentage
<	Less than
±	Plus-minus sign
a.m.	Ante meridiem
B1	Thiamine
B2	Riboflavin
B6	Pyridoxine
bpm	Beats per minute
BRUMS	Brunel mood scale
cm	Centimetres
ED	Energy drink
ES	Effect sizes
FAS	Felt arousal scale
FS	Feeling scale
g	Gram
hr	Hour
HR	Heart rate
HREC	Human research ethics committee
IBM	International Business Machines Corporation
ID	Identification
IPAQ-M	International physical activity questionnaire
JAKIM	Department of Islamic Development Malaysia
kg	Kilogram
m	Metres
max	Maximum

MET	Metabolic equivalent
mg	Milligram
min	Minute
mL	Millilitre
msec	Millisecond
N	Total sample size
NY	New York
p.m.	Post meridiem
PAR-Q	Physical activity readiness questionnaire
PD	Placebo
RPE	Rating of perceived exertion
rpm	Revolutions per minute
s	Second
SD	Standard deviation
SOP	Standard operating procedure
Tmax	Time taken to reach the maximum concentration
UK	United kingdom
USM	Universiti Sains Malaysia
vs	Versus
W	Watt
WHO	World Health Organization

**KESAN PENGGUNAAN MINUMAN TENAGA TERHADAP PRESTASI
FIZIKAL DAN TINDAK BALAS PERSEPSI DALAM KALANGAN PELAJAR
UNIVERSITI YANG TIDAK AKTIF SECARA FIZIKAL**

ABSTRAK

Penggunaan minuman tenaga (contohnya RedBull, Livita, Monster Energy) telah menunjukkan peningkatan yang besar sejak beberapa dekad yang lalu, terutama dalam kalangan pelajar universiti. Penyelidik telah melaporkan peningkatan prestasi senaman (daya tahan, ketangkasan) dengan penggunaan minuman tenaga dalam kalangan atlet atau individu yang aktif secara fizikal, namun penemuan ini masih kurang jelas. Pada masa ini, kesan minuman tenaga dalam kalangan individu yang tidak aktif secara fizikal masih tidak jelas. Tujuan kajian ini adalah untuk mengkaji kesan minuman tenaga terhadap prestasi fizikal (iaitu, prestasi ketangkasan, masa tindak balas, prestasi maksimum jangka pendek) dan tindak balas persepsi (iaitu, keadaan mood, kadar tanggapan daya usaha (RPE), degupan jantung (HR), tindak balas afektif) dalam kalangan pelajar universiti yang tidak aktif secara fizikal. Reka bentuk kajian yang rawak, bersilang dan pengukuran berulang dilaksanakan dalam kajian ini. Seramai sebelas pelajar universiti yang tidak aktif secara fizikal ($N = 11$, 4 lelaki dan 7 perempuan, umur = 22.82 ± 0.98 tahun; tinggi = 164.13 ± 4.81 cm; jisim badan = 59.83 ± 13.88 kg; jumlah MET-minit / minggu = 578.73 ± 161.20) mengambil bahagian dalam kajian eksperimen ini. Semua peserta menjalani dua keadaan eksperimen: keadaan minuman tenaga (ED) dan keadaan plasebo (PD). Peserta melengkapkan soal selidik Brunel Mood Scale (BRUMS) sebelum dan selepas kajian eksperimen ini. Selepas itu, para peserta membuat pemanasan badan dengan berlari di atas 'treadmill' selama 3 minit pada kadar yang dipilih sendiri diikuti dengan ujian-t ketangkasan, ujian ringkas masa reaksi ketukan visual, dan ujian Wingate 30-s.

Tempoh rehat selama 3 minit diberikan antara setiap ujian. Tindak balas persepsi, iaitu, RPE, HR dan tindak balas afektif diukur dan dihitung sejurus selepas ujian-ujian ini. Data dianalisis menggunakan analisis pengukuran varians satu arah (ANOVA) untuk memeriksa perbezaan merentasi pemboleh ubah bersandar. Penemuan kajian ini menunjukkan penurunan tahap keletihan yang signifikan dan persepsi skor kemurungan pada ED berbanding dengan PD dalam BRUMS. Kajian ini juga menunjukkan bahawa tindak balas afektif yang diukur melalui skala perasaan (FS) adalah bertambah baik secara signifikan dengan pengambilan ED dalam purata perubahan skor FS semasa ujian-t ketangkasan ($p = 0.001$) sementara PD menghasilkan peningkatan yang signifikan ($p = 0.02$) pada perubahan skor FS semasa ujian 30s-Wingate. Juga, penggunaan ED menghasilkan magnitud yang lebih besar ($p = 0.003$) dalam perubahan min skor skala rangsangan rasa (FAS) semasa ujian ketangkasan berbanding PD. Di samping itu, kajian ini mendedahkan bahawa ED membawa kepada perubahan RPE yang baik ($p = 0.039$) daripada PD dalam kalangan individu yang tidak aktif secara fizikal semasa melakukan ujian masa reaksi ketukan. Akhirnya, tidak ada perbezaan yang signifikan dalam pemboleh ubah yang lain (iaitu, ujian-t ketangkasan, ujian masa reaksi ketukan, dan ujian Wingate 30-s) berkait dengan kesan ergogenik minuman tenaga. Oleh itu, sebagai kesimpulan, kajian ini menunjukkan bahawa pengambilan ED mempunyai kesan positif untuk mengurangkan keadaan mood negatif iaitu skor keletihan dan kemurungan antara pelajar universiti yang tidak aktif secara fizikal. Dari segi tindak balas afektif (FS dan FAS), penggunaan ED telah menunjukkan beberapa peningkatan yang ketara daripada PD. Kajian dan penyelidikan lebih lanjut diperlukan untuk pemahaman yang mendalam tentang manfaat minuman tenaga yang mengandungi taurin terhadap prestasi fizikal dan tindak balas persepsi dalam kalangan individu yang tidak aktif secara fizikal.

EFFECTS OF ENERGY DRINK CONSUMPTION ON PHYSICAL PERFORMANCE AND PERCEPTUAL RESPONSES AMONG PHYSICALLY INACTIVE UNIVERSITY STUDENTS

ABSTRACT

Consumption of energy drinks (e.g. RedBull, Livita, Monster Energy) has been increasing during the past few decades, especially among University students. Researchers have reported improvements in exercise performance (i.e., endurance, agility) with the use of energy drink among athletes or physically active individuals, although these findings are equivocal. Currently, the effects of energy drink among physically inactive individuals is unclear. The aim of the present study was to examine the effects of energy drink on physical performance (i.e., agility performance, reaction times, short-term maximal performance) and perceptual responses (i.e., mood state, rating of perceived exertion (RPE), heart rate (HR), affect responses) in physically inactive university students. A randomized, repeated measures cross-over design was implemented in this study. A total of eleven physically inactive students ($N = 11$, 4 males and 7 females, age = 22.82 ± 0.98 years; height = 164.13 ± 4.81 cm; body-mass = 59.83 ± 13.88 kg; total MET-minutes/week = 578.73 ± 161.20) participated in this experimental study. All participants underwent two experimental conditions: energy drink (ED) condition and placebo condition (PD). Participants completed the Brunel Mood Scale (BRUMS) questionnaire before and after the exercise protocols. Subsequently, participants performed a 3-min warm-up by running on a treadmill at self-selected paced followed by agility t-test, simple visual tap reaction time test, and the 30-s Wingate test. A 3-min rest gap was given between each exercise trial. Perceptual responses, namely, RPE, HR and affect responses were measured and calculated immediately after these tests. Data were analysed using one-way repeated

measures analyses of variance (ANOVA) to examine differences across dependent variables. Findings of the present study showed significant reductions in level of fatigue and the perception of depression scores in ED compared to PD in BRUMS. This study also indicates that affect response measured via feeling scale (FS) was significantly improved with the ingestion of ED in mean changes of score of FS during agility t-test ($p = 0.001$) while PD elicited a significant improvement ($p = 0.02$) in the mean changes of score of FS during 30s-Wingate test. Also, consumption of ED elicited a greater magnitude ($p = 0.003$) in the mean changes of score of felt arousal scale (FAS) during agility t-test compared to PD. In addition, this study revealed that ED leads to favourable changes of RPE ($p = 0.039$) than in PD among physically inactive individuals while performing tap reaction time test. Finally, there were no significant differences in other variables (i.e., agility t-test, tap reaction time test, and 30-s Wingate test) associated with the ergogenic effects of energy drink. Thus, in conclusion, this study demonstrated that ED ingestion has a positive effect on reducing the negative mood states which are fatigue and depression scores among physically inactive university students. In terms of affect responses (FS and FAS), ED consumption has showed several significant improvements than that of PD. Further studies and research are warranted for in depth understanding of the benefits of taurine-containing energy drinks on physical performance and perceptual responses among physically inactive individuals.

CHAPTER 1 : INTRODUCTION

1.1 BACKGROUND OF THE STUDY

Energy drinks are a type of beverage used by consumers (e.g. athletes, children, and elderly) to provide as an energy boost. Energy drinks which are usually carbonated and contain significant quantities of sugar and caffeine as well as blends of herbal extracts, B vitamins, and amino acids which are typically assumed to provide increased energy and significant improvements in cognition (Malinauskas et al., 2007). Previous evidences have reported that the market and degree of consumption of energy drinks is inclining every year particularly among university students (Malinauskas et al., 2007). An explanation for the popularity of energy drinks among university students may be attributed to believe that energy drinks could give an 'energy boost' in the form of increased alertness, visual information processing, attention, physical performance, and to combat fatigue. Despite all of these notions, the effects of energy drinks on physical and psychological performances remain controversial especially in physically inactive university students.

There are available evidences to indicate the benefits of energy drinks to enhance physical and psychological performances in adults (Alford et al., 2001). For example, Alford and colleagues (2001) has reported that energy drinks improved aerobic endurance (maintaining 65-75% max. heart rate) and anaerobic performance (maintaining max. speed) on cycle ergometers in physically active adults. Moreover, the author also indicated that the ingestion of energy drinks elicited enhancement in mental performance including choice reaction time, concentration, and memory. Despite an emerging body of evidence to indicate the ergogenic aids of energy drink on physical and mental performances, these studies are limited to physically active individuals and athletes. Likewise, Hahn and colleagues (2018) claimed that there were

no beneficial effects of caffeine-containing ED on vertical jumping and repeated sprinting (i.e., measures of mean and peak anaerobic power) on recreationally active men. According to Gwacham and Wagner (2012), they have observed no ergogenic effect of caffeine-aurine ED on repeated sprinting (i.e., 6 × 35-s with 10-s rest intervals) as well among the American college football players. In addition, a single study conducted by Al-Fares and colleagues (2015) has reported that no significant difference between energy drink and placebo on time to exhaustion performance, VO₂max, blood pressure and heart rate in untrained women, indicating that energy drink may not elicit beneficial effect in this group. However, the author did not report details on participants' physical activity or exercise experience. Furthermore, this previous study only investigated the effects of energy drink on time to exhaustion and it is not possible to extrapolate this evidence to other physical performances parameters such as agility, Wingate tests and reaction time as well as mental performance (e.g. RPE and mood). Consequently, the beneficial effects of energy drinks on physical and mental performances are remain unclear among inactive population.

Therefore, the purpose of this present study was to evaluate the influence of energy drinks on the physical performance (i.e. agility, anaerobic performance, reaction time) and psychological responses (i.e. mood, affective responses and perceived exertion) among physically inactive university students.

1.2 PROBLEM STATEMENT

There is evidence that demonstrates the effects of energy drinks on physical performance and psychological performance, but the relevant research is limited to physical active individual and athletes, and fewer studies have investigated the impact of energy drink in physically inactive adults. The present study is important so that physically inactive individuals may adhere more to physical activity or exercise with the assist of energy drinks. Documentation of this evidence is necessary to once and for all determine the true potential, if any, of the effect of energy drinks on physiological and psychological parameters to facilitate the improvement in physical performance while promoting the importance of exercise among university students.

1.3 OBJECTIVES OF THE STUDY

1.3.1 General Objective

To determine the effects of energy drinks (ED) consumption on physical performance and perceptual responses among physically inactive university students.

1.3.2 Specific Objectives

1. To investigate the effects of ED on physical performance (e.g. agility, anaerobic performance, and reaction time) among physically inactive students.
2. To investigate the effects of ED on perceptual responses (e.g. mood, affective responses, and perceived exertion) among physically inactive students.

1.4 RESEARCH QUESTIONS

1. Is there any significant difference between consuming ED and carbonated drinking water (placebo) on physical performance (e.g. agility, reaction time, and anaerobic performance) among physically inactive university students?
2. Is there any significant difference between consuming ED and placebo on perceptual responses (e.g. mood, affective responses (FS and FAS), and perceived exertion) among physically inactive university students?

1.5 HYPOTHESES OF THE STUDY

Ho1 : There is no significant difference between consuming ED and placebo on physical performance among physically inactive students.

Ha1 : There is a significant difference between consuming ED and placebo on physical performance among physically inactive students.

Ho2 : There is no significant difference between consuming ED and placebo on perceptual responses among physically inactive students.

Ha2 : There is a significant difference between consuming ED and placebo on perceptual responses among physically inactive students.

1.6 SIGNIFICANCE OF THE STUDY

Both physical performance (agility, anaerobic performance, reaction time) and perceptual responses (mood, affective responses and perceived exertion) are important across variety of groups (e.g. adult, athletes, older adults, children) including physically inactive individuals as these two factors reflect the individual's levels of physical fitness and motivation towards exercise, respectively. Indeed, these two factors have been shown to facilitate in enhancement of multiple health benefits (physical performance) and future exercise engagement (perceptual responses). Consequently, it is crucial to truly understand one's own physical, perceptual capabilities and limits. Thus, this proposed study will potentially provide valuable information and guidelines regarding the effects of energy drink on physical performance and perceptual responses especially in physically inactive groups.

1.7 OPERATIONAL DEFINITIONS

Affective responses :

General psychological state of an individual, including but not limited to emotions and mood, within a given situation. It used to describe an individual's subjective experience (i.e., intrapersonal, or experimental core) of all valence responses (positive and negative dimensions).

Agility t-test :

The agility t-test is a test that is commonly used to assess athletes or individuals' ability to move forward, lateral, and backward, appropriate to a wide range of sports.

Tap reaction time test :

An online virtual tap reaction time test to assess a person's quickness to react to a stimulus. For example, by responding to a change in screen colour by tapping the screen as fast as possible.

Taurine :

Taurine is a normal metabolite in humans that is involved in the modulation of neuronal excitability, membrane stabilization, production of bile salts, and the detoxification of certain xenobiotics. It is estimated that the daily intake of taurine in humans is between 40 and 400mg.

Perceived exertion response :

Rating of perceived exertion (RPE) is a recognized marker of intensity during exercise. It is based on the physical sensations an individual experiences during physical activity, including increased heart rate, increased respiration or breathing rate, increased sweating, and muscle fatigue.

Physically inactive individuals :

Individuals who do not meet the criteria of total physical activity of at least 600 MET-minutes/week is classified as physically inactive.

30s-Wingate test :

Test of maximal anaerobic power output during 30 seconds of all-out exercise on a leg-cycle ergometer; a measure of maximal power output and capacity of immediate and short-term (glycolytic) energy systems.

CHAPTER 2 : REVIEW OF LITERATURE

2.1 ENERGY DRINK

According to Sullivan, R. (n.d.), energy drinks are beverages that are marketed explicitly to enhance performance and boost energy in which they usually contain various quantities of sugar, caffeine, taurine, ginseng, guarana and other proprietary ingredients. In addition, energy drinks are popular among youth and are regularly consumed of approximately 31% of 12 to 17-year-old and 34% of 18 to 24-year-old (Sullivan, n.d). The other reason behind why people of all ages will buy and consume energy drinks not just only to relieve fatigue, increase brain function and improve one's mental alertness, but also due to the convenience and availability of the energy drinks as it can be purchased anywhere such as grocery stores, shopping malls, convenience stores, vending machines and also gas stations.

The difference between energy drink and sport drinks is that energy drinks typically contain stimulants (e.g. caffeine and guarana) and amino acids (e.g. taurine), as well as added sugars and often vitamins, minerals and other nutrients (American Academy of Pediatrics, 2011; Higgins et al., 2010). Besides that, energy drinks are marketed as a means of boosting energy, decreasing feelings of tiredness and enhancing alertness (American Academy of Pediatrics, 2011; Higgins et al., 2010). On the other hand, despite their added sugar content, sports drinks usually contain electrolytes, minerals, vitamins and other nutrients (American Academy of Pediatrics, 2011; Coombes & Hamilton, 2000) and are marketed as a means of improving athletic performance by replacing the electrolytes and fluid lost in sweat during and after intense physical activity (American Academy of Pediatrics, 2011; Coombes & Hamilton, 2000). Since not much studies have shown the effects of energy drinks among inactive individuals, hopefully this research study is able to bring some valuable information and

guidelines on the effects of energy drinks on physical performance and perceptual responses among physically inactive university students.

2.2 BENEFITS OF ENERGY DRINKS ON PHYSICAL PERFORMANCE

Physical performance is also known as physical function whereby one's ability to carry out activities that require physical actions, ranging from self-care (e.g. activities of daily living) to more complicated activities that require a combination of motor skills, often with a social component or within a social context (Lummel et al., 2015). Judging from outcomes obtained in Petrelli et al. (2018) study, energy drinks are able to provide an additional burst of energy in a short-time period, more noticeable in trained subjects than untrained. For this reason, to achieve benefits from the intake of an energy drink, short and low-intensity activities are most favored. Throughout the study, it is notable that there are effects of energy drinks regarding physical performance on trained individuals, however these studies are limited especially for physical inactive population.

As mentioned sports activities most suited to exploiting the effects of energy drinks claim to give are short-term and low intensity activities. In this study, participant's agility, anaerobic performance and reaction time will be evaluated to observe the effects of energy drinks. Agility has been defined as the ability to maintain a controlled body position and rapidly change direction without a loss of balance, body control, or speed (Roozen, 2004; Miller et al., 2006). A study done by Coso et al. (2014) reported that the time to complete the agility test was significantly reduced with the consumption of energy drink (10.8 ± 0.7 vs 10.3 ± 0.4 s, $P < 0.05$) in male volleyball players. Another reason of performing agility t-test because it is relatively simple to administer because it requires minimal equipment, materials and preparation.

On the other hand, tap reaction time test (MathIsFun, 2016) will be assessed as it will be used to evaluate the quick response and required full focus from the participants by tapping the changed colour dot on screen. Alford et al. (2001) saw a positive effect on reaction time (a decrease of 88.7 msec) with the consumption of energy drinks. A result of quick reaction test is important as it means that the brain and spinal cord can quickly send the messages, information or signals to your bones, muscles and joints in order to function well or prepare for any movements. In this study, participants used their own smartphones to assess tap reaction time test to increase the reliability of test and to prevent the risk of COVID-19.

Although several studies have investigated the effects of energy drinks on aerobic performance, there is as yet only limited and inconclusive data about their impact on short-term maximal performance (Hahn et al., 2018). Main findings from the study done by Chtourou et al. (2019) were that energy drink increases peak power (+0.93 W.kg⁻¹) and mean power (+0.87 W.kg⁻¹) during the 30-s Wingate test. Wingate anaerobic test (generally called 'Wingate Test') first presented by Ayalon et al., (1974), was derived from the test previously proposed by Cumming (1974). Thereafter, Bar-Or (1978; 1987) published comprehensive studies of the Wingate test and its applications. In agreement with the results from Chtourou et al. (2019), Alford et al. (2001) reported that energy drink had a positive effect on short-term maximal performance during the Wingate test.

It is very important to note that although the above-mentioned studies have identified positive effects of energy drinks on exercise performance, other researches have documented no significant effects or detrimental health consequences. Al-fares et al. (2015) in a single blind placebo-controlled study recently evaluated the effects of energy drinks on exercise performance in 32 untrained healthy females. The authors found that ingestion of energy drinks before exercise did not enhance the indices of

physical performance, which included time to exhaustion, maximum oxygen consumption, blood pressure, heart rate, and capillary oxygen saturation. This finding may indicate that variations in participants' background (e.g. physical activity levels and exercise experience) may be contributed to the inconsistency of the findings.

2.3 BENEFITS OF ENERGY DRINKS ON PERCEPTUAL RESPONSE

Perceptual is the ability to interpret or differentiate objects, being aware of something through the senses. Secondly, perceptual is an important process as it allows us to be able to focus our attention on more salient events or objects and, in addition, allows us to categorize such events or objects so that they fit into our own conceptual map of the environment (Bruner & Postman, 1949). Studies of relationships between energy drinks and psychological variables are also inconclusive to date; however, some studies have reported positive effects on subjective alertness, mental focus, energy, and fatigue tolerance (Hahn et al., 2018; Alford et al., 2001; Hoffman et al., 2009; Brunye et al., 2010; Jagim et al., 2016). A study done by Hahn and colleagues (2018) described that there was a significant reduction of perceived fatigue during repeated sprinting among recreationally active men. According to Hoffman and colleagues (2009), it had been reported significant improvements in athletes' focus (+0.5 arbitrary units, AU) and energy (+0.4 AU) after ingestion of caffeine-containing EDs compared to placebo (PL). Likewise, Wesnes et al. (2017) demonstrated significant improvement in the attentional capacity, vigilance, and numeric and spatial working memory of healthy young adults after ingesting energy drinks. However, no significant changes in mood state were seen. In contrast with this, Petrelli et al. (2018) reported significant reductions of anxiety and depression after ingestion of energy drinks consumption compared to placebo group. From the previous studies mentioned above, ingestion of ED might help to improve one's alertness, mental focus, fatigue tolerance and reduce negative mood scores such as anxiety and depression.

The intake of energy drinks has also caused an increase in somatic-affective factors, decreasing cognitive ones and the perception of depression. To determine each participant's perceptual responses, two methods were used in this study which are Brunel Mood Scale (BRUMS) and also affective response. Subsequent research that has tested the factor structure of the BRUMS has been supported in specific-sports such as wakeboarding (Fazackerley et al., 2003) and among special populations. The purpose of BRUMS is to provide a quick assessment of mood states for adolescents and adults. The BRUMS has shown predictive validity (Lane & Chappell, 2001; Lane et al., 2001), and be responsive to the effects of exercise (Lane & Lovejoy, 2001). Further, the BRUMS has been used in applied settings in the screening of athletes (Galambos et al., 2005) and normative data for use with athletes has been developed for use with UK athletes (Terry & Lane, 2010). However, there are no information about the relationship between mood and physical performance exists in inactive population.

Whereas for affective response that commonly measured using a single item scale known as Feeling Scale, was used as a measure of “basic” or “core” affective valence (pleasure–displeasure). Specifically, affective responses can be defined as a pleasure and displeasure feelings during exercise. In contrast to the persistent general belief that exercise is enjoyable for everyone, strong individual differences are found in the affective responses during and after exercise. Whereas some individuals indeed report an increase in pleasure or no change, others report reduced pleasure or negative changes in affect (Ekkekakis et al., 2005; Ekkekakis et al., 2011; Van Landuyt et al., 2000; Welch et al., 2007). If the affective response is on balance positive, people are more likely to maintain the behaviour and become regular exercisers. However, if the net affective response is not favourable, people are at risk of dropping out and become non-exercisers. Indeed, evidence has shown that affective responses during

exercise may influence future attitudes towards PA behaviour in adult (Schneider et al., 2009).

CHAPTER 3 : METHODOLOGY

3.1 STUDY DESIGN

This study employed a repeated measures cross-over design, whereby participants completed two experimental trials involving energy drink (ED) and placebo (PD) conditions with 7 days of washout period. Duration of participants' involvement in this study was up to four weeks. Participants were recruited based on the inclusion and exclusion criteria that were set at the beginning of the study. All outcome measurements were taken twice; before (first session) and after (second session) the trial. This study procedures were approved (Appendix A) by Human Research Ethics Committee (HREC), Health Campus of Universiti Sains Malaysia (USM/JEPEM/21010022). This study had no conflict of interest.

3.2 STUDY LOCATION

The data collection was conducted at Exercise and Sport Science Laboratory of School of Health Sciences, Health Campus, Universiti Sains Malaysia (USM), Kubang Kerian, Kelantan. Anthropometric measurements such as height, weight, body mass index (BMI) and the exercise protocols were collected and performed at the laboratory of School of Health Sciences in USM.

3.3 SAMPLE SIZE CALCULATION

Sample size was calculated by using G*Power version 3.1.9.2 (Faul et al., 2007). Based on the results of previous study (Chtourou et al., 2019), effect sizes (ES) were estimated to be medium effect ($ES = 0.45-0.54$) across all outcome variables (e.g. agility, anaerobic performance, reaction time and mood responses). Data were analysed using a one-way repeated measures ANOVA to examine differences in the outcome variables between conditions (ED and PD). To reach the desired statistical power and in order to attribute observed differences to factors other than chance alone, a minimum sample of 12 participants will be required using a power of 0.8, an alpha of 0.05 and an effect size, F , of 0.30 (medium).

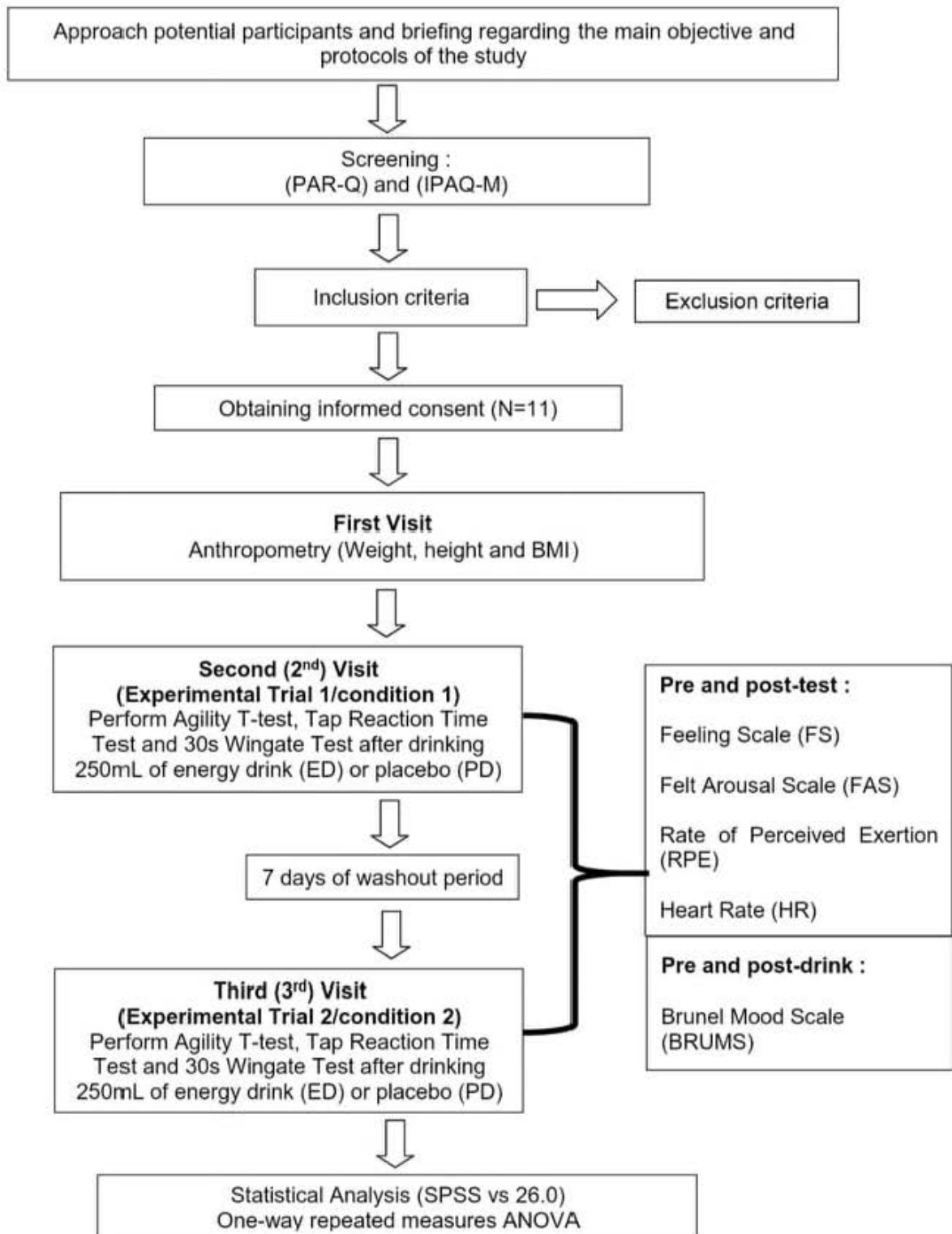


Figure 3.1: Flow chart of the study procedures.

3.4 PARTICIPANTS

A total of twelve healthy physically inactive males and females were recruited among students of Universiti Sains Malaysia via advertisement (Appendix B) posted in social media (e.g. Whatsapp, Instagram). However, results were presented for eleven participants, as one participant dropped out for personal reasons unrelated to this study. The participants gave informed consent form after being advised of all the possible risks and discomforts associated with the procedures used in the study. The inclusion and exclusion criteria were as follows :

Table 3.1 : Inclusion and exclusion criteria.

Inclusion Criteria	Exclusion Criteria
Male and female students of USM	Having musculoskeletal injuries
Non regular caffeine users (consuming less than one cup of coffee or caffeine equivalent daily)	Intake of any medications or dietary supplements known to influence blood glucose concentrations
Aged between 18 to 30 years old	Diagnosed with any chronic metabolic disease
Healthy individuals	
Physically inactive (individuals who do not meet the criteria of total physical activity of at least 600 MET-minutes/week is classified as physically inactive)	

3.5 STUDY OVERVIEW

During the recruitment process, potential participants were approached by researcher and they were thoroughly explained regarding the objectives, procedures, possible harm/risk and benefits of the research study. Potential participants were requested to complete a Physical Activity Readiness Questionnaire (PAR-Q; Jeal 2020) (Appendix C) before commencing experimental tests to prevent any unwanted health risks, to assess their level of physical fitness and their ability to engage in any physical activity. Followed by completing the Malay version of the International Physical Activity Questionnaire (IPAQ; Lee, Macfarlane, Lam & Stewart, 2011) (Appendix D), which was used to measure habitual physical activity levels of the participants. Those who were voluntarily to participate and met the study criteria were successfully enrolled for the study. All participants completed the informed consent form (Appendix E) and the participant information collection form (Appendix F).

This study required three experimental trials in the laboratory, separated by a minimum 7-d rest period (wash out period), and incorporated a within-measures design. The first visit was to measure anthropometric variables and familiarize participants with the measurement scales. This was followed by two experimental visits involving with energy drink (ED) and placebo (PD) conditions, the order of which was counterbalanced to control for any order effect. Lottery method was used by the researcher as simple random sampling to determine which participants received ED or PD on experimental trial 1 and 2.

Researcher and the use of laboratory were available from 10.00a.m. until 4.30p.m. so that participants can come during their free time and also to obey the rule of standard operating procedure (SOP) by not having a mass of students coming into the laboratory all at once. Besides that, researcher ensured that every participant

wears a mask and fills up the form to get a permission pass prior entering the laboratory.

3.6 MEASUREMENT INSTRUMENTS AND PROCEDURES

3.6.1 FIRST VISIT: ANTHROPOMETRIC AND PHYSICAL ACTIVITY

The first session comprised of fifteen to thirty minutes of briefing up/familiarization, signing consent form, PAR-Q form, IPAQ-M form, participants' ID and for pre-measurement. Upon arrival, participants' body weight and height were measured by a body composition analyser (Omron, Japan) and a stadiometer (Seca, China) respectively. Body mass and stature were measured to the nearest 0.1 kg and 0.1 cm, respectively (the participants were shoeless and wear light clothing). Then, calculation of body mass index was done as follows :

$$\text{BMI (kg.m}^{-2}\text{)} = \text{weight (kg)} / \text{height (m)} \times \text{height (m)}$$

Participants completed Malay versions of the International Physical Activity Questionnaire short form (IPAQ-M; Lee, Macfarlane, Lam & Stewart, 2011) to determine habitual PA levels. IPAQ has been recommended as a cost-effective method to assess physical activity (Lee et al., 2011). IPAQ-M can be divided into three levels of categorical score that consists of Category 1 (Inactive; <600 MET-min/week), Category 2 (moderately active; <3000 MET-minutes/week) and Category 3 (health-enhancing physical activity (HEPA); >3000 MET-minutes/week).

3.6.2 SECOND AND THIRD VISITS: EXPERIMENTAL PROTOCOLS

After that, each participant was required to visit the Exercise and Sport Science laboratory of School of Health Sciences for two experimental sessions, drinking an energy drink (ED) and carbonated drinking water which serve as placebo (PD).

Participants were asked to abstain from caffeine, taurine, over-the-counter medications, and any herbal supplements for 24 hr period before each test session. Beverages were prepared and chilled in a refrigerator by the lab assistant who took no part in the test session or data analysis.

Next, they were asked to drink the fluid quickly 60 min prior their test session and not to discuss or compare tastes or to make any assumption about what they have consumed. The interval of 60 min was chosen as being an optimal for a complete respective drinks absorption and thus enabling the peaking of active component concentration. As peak plasma concentrations of energy drink after oral administration have been reported to occur at a T_{max} of 30-120 min (Carrillo & Benitez, 2000). Participants were fully supervised by researcher to ensure that they drink the entire quantity of fluid and no leftover. After consumption, participants were required to complete all the physical performance test: 1) agility test; 2) tap reaction time test; and 3) 30s-Wingate test. All sessions were arranged in the same day to avoid any time of day effects. Following that, participants performed and completed all the exercise protocols as mentioned for one session each week for a total of two weeks. The two definitive test sessions were separated by an interval of seven days to allow sufficient recovery between tests and to ensure active component washout (Chtourou et al., 2019). To avoid identification, ED or PD were ingested by each participant (i.e., 250 mL) from the two unmarked and non-transparent water bottles in the presence of a researcher. Moreover, participants were not allowed to open the water bottle to drink or to peek at the given beverage.

The ED drink (i.e., 250 mL) contained 1000 mg of taurine, 16.5 g for total carbohydrates and sugar, 0 g for both protein and fat content, 20 mg of niacin, 2 mg of vitamin B1, 2.9 mg of vitamin B2, 3.6 mg of vitamin B6, contains a tiny amount of inositol, citric acid, natrium benzoate, pyridoxine added with artificial flavors (Spargo,

2020). Possible minor side effects of consuming ED drink including insomnia, nervousness, increased heart rate, sugar crashes and weight gain (Spargo, 2020). However, there were no serious harm towards the participants since they have submitted their PAR-Q forms before commencing any experimental tests. Next, commercialize energy drink which was available in the market were used for the purpose of this study. Therefore, there was no issues related to the Halal certification. For the placebo drink, commercial sparkling water which was also available in Malaysia market were used. ED drink was Halal certified according to Nasrudin and his colleagues (2011). Meanwhile the PD drink do not contain any caffeine, taurine, sugar, carbohydrate, fat, protein, calcium, iron, vitamin A nor C. PD drink have no side effects as it is made with real fruit flavors, natural spring water and refreshing bubbles. It was Halal certified by the Department of Islamic Development Malaysia (JAKIM) and other relevant authorised Islamic certification bodies in accordance with the Halalan Toyiyban standards and the Islamic Syariah Law requirements (Fraser & Neave Holdings Bhd, 2021). Furthermore, both ED and PD drinks were bought and checked prior tests.

As shown in the Table 3.2, after consuming the given beverages (i.e., 250 mL) for an hour, participants started off with a warming up session by running on a treadmill for 3 minutes, following their own pace. 3 trials of agility t-test were completed and participants got to rest for 3 minutes. Participants were explained by the researcher on how to perform the agility t-test properly to avoid unnecessary injuries (please refer to 3.6.3). Followed by performing the tap reaction time test (please refer to 3.6.4), participants completed all 5 trials and got the same rest of 3 minutes. They were briefed as well by the researcher about the details and purpose of the reaction time test. Lastly, participants completed the 30s-Wingate test (please refer to 3.6.5) as the last experimental test with just one trial. Participant's heart rate (HR - Appendix L), rating of perceived exertion (RPE - Appendix G), Feeling Scale (FS - Appendix I) and Felt

Arousal Scale (FAS - Appendix J) were recorded immediately after each exercise session. BRUMS Questionnaire (Appendix H) were filled by participants before and after the exercise protocols.. All the details related to the measurements are explained in the following section. Participants were taught and shown how to perform the exercise correctly to reduce the risk of musculoskeletal or orthopaedic injuries. In addition, all the exercise sessions began with a warm-up on a treadmill for 3 minutes and ended with a cool-down for 3 minutes. All the exercise sessions were conducted at Exercise and Sport Science Laboratory of School of Health Sciences in USM and each session were fully supervised by the researcher.

At the end of the study, eleven participants completed the intervention successfully. The cross-over intervention study was carried out 1 time per week for 2 weeks including the first briefing session which were a total of 3 sessions. The research protocol including trial testing were conducted on Sunday, Monday and Wednesday. All the data obtained were recorded in the data collection forms (Appendix L).

Table 3.2 : Energy drink and placebo intervention programme (carried out twice with 7 days of washout period).

Consuming 250mL energy drink (ED) or placebo (PD), fill in BRUMS *60 min prior experimental tests*
Warm-up for 3 minutes (running on a treadmill)
Agility T-Test (3 trials)
Rest for 3 mins while filling in heart rate, RPE, FAS and FS
Tap Reaction Time Test (5 trials)
Rest for 3 mins while filling in heart rate, RPE, FAS and FS
30s Wingate Test

(1 trial)
Rest for 3 mins while filling in heart rate, RPE, FAS, FS and BRUMS
End of Session (Repeat all protocols on the second week)

3.6.3 AGILITY T-TEST

The t-test (Semenick, 1990) was administered using the protocol outlined by Lockie et al. (2016) to measure the ability to turn in different directions. Participants began the test with both feet behind the starting line. At their own discretion, each participant performed sprint forward 5m towards the middle cone and touch the base of a cone using their right hand. They then shuffled to the left (2.5m) and touched the base of the left cone again with their left hand. Participants then shuffled to the right side (5m) and touched the base of the right cone with their right hand. They then shuffled back again to the middle cone (2.5m) and touched the base of the middle cone. Lastly, participants ran backward, passing the finishing line at the starting line to complete the test. All participants were required to perform this test for three times.

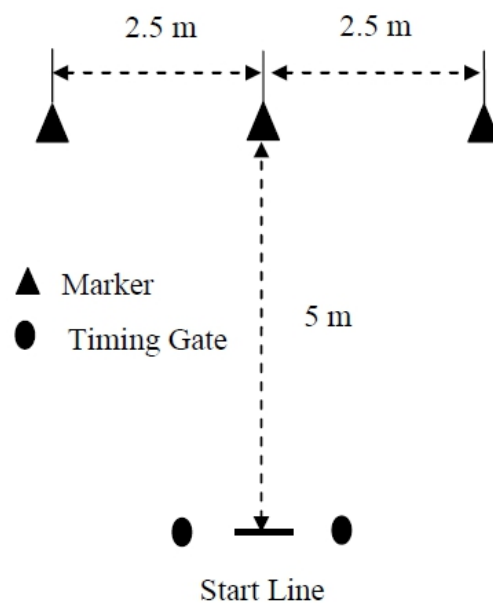


Figure 3.2: Layout of the T-test. (Modified from Lockie et al., 2016).

3.6.4 REACTION TIME/RESPONSES EVALUATION

Participants' reaction time were observed via online (<https://www.mathsisfun.com/games/reaction-time.html>) using the Tap Reaction Time Test (MathIsFun, 2016). All participants responded to a change in the screen colour by tapping the screen as quickly as possible. The speed for each attempt was recorded over five attempts in second (s) and average value were collected.

3.6.5 30s-WINGATE TEST

Wingate test consists in pedalling with maximal (all-out) effort for 30 seconds against a constant braking force (7.5% BW for a Monark ergometer). The mean pedal rate is measured for each 5-second interval and for the Monark Ergonomic 839 E Cycle Ergometer, mean power outputs corresponding to these intervals are given by the product of braking force and mean pedal rate (Driss & Vandewalle, 2013). Before the test began, participants pedalled at low pedal rate with a low resistance for a few minutes. With sufficient amount of warm up, all participants then performed the 30s-Wingate test by pedalling a maximal (all-out) effort for 30 seconds, then followed by pedalling slowly to make sure there was enough blood flowing through the activated muscles (legs). Peak power output (PP), mean power output (MP) over the 30 seconds of the whole test and the maximum speed (rpm) of participants were computed.

3.6.6 AFFECTIVE RESPONSES

The Feeling Scale (FS; Hardy and Rejeski, 1989) was used as a measure of "basic" or "core" affective valence (pleasure–displeasure). Participants responded to how they feel on an 11-point bipolar scale ranging from "Very Good" (+5) to "Very Bad" (-5). Perceived activation levels was measured using the single-item felt arousal scale (FAS; Svebak & Murgatroyd, 1985). Participants were asked to rate themselves on a 6-point scale ranging from 1 'low arousal' to 6 'high arousal'. FS and FAS exhibited

correlations ranging from 0.41 to 0.59 and 0.47 to 0.65, respectively, with the Affect Grid (Russell et al., 1989), indicative of convergent validity with similar established measures (Van Landuyt et al., 2000). All FS and FAS scores were calculated using the changes of score between post and pre of FS and FAS. Participants responded to the FS and FAS 5 minutes before test/exercise, and immediately after test/exercise. Participants were also given standardized verbal instructions on how to use the scales before undertaking the incremental test and at the start of the exercise session using the below sentences:

- i. **Feeling Scale:** While participating in exercise, it is quite common to experience changes in mood. Some individuals find exercise pleasant, whereas others find it to be unpleasant. Additionally, feeling may fluctuate across time. That is, one might feel good and bad a number of times during exercise. How does above scenario make you feel during the exercise?
- ii. **Felt arousal scale:** Estimate here how aroused you actually feel. By “arousal” we meant how “worked-up” you feel. You might experience high arousal in one of a variety of ways, for example as excitement or anxiety or anger. Low arousal might also be experienced by you in one of a number of different ways, for example as relaxation or boredom or calmness.

3.6.7 BRUNEL MOOD SCALE (BRUMS)

Mood states of participants were measured using the Malaysian Brunel Mood Scale (BRUMS; Hashim, Zulkifli, & Hanafi, 2010). It contained 24-item questionnaires of simple mood descriptors such as angry, nervous, unhappy, and energetic. BRUMS had six subscales: anger, confusion, depression, fatigue, tension, and vigour. Responses were recorded using 5-point likert scale, where 0 (Not at all), 1 (A little), 2 (Moderately), 3 (Quite a bit) and 4 (Extremely). BRUMS questionnaire had been